30. (Twice Amended) A substrate having a plurality of probes, wherein said probes are fluorescently labeled by incorporation of at least one nucleotide analog, the labeled probe providing a detectable first level of fluorescence, and when hybridized to a complementary target having no nucleotide analogs incorporated therein providing a second level of fluorescence, wherein the second level is greater than zero, said levels of fluorescence being derived from excitation of said at least one nucleotide analog.

C6

31. (Amended) A substrate having a surface area, the surface area comprising attached labeled probe molecules, said probe further comprising a fluorescent label, said fluorescent label including at least one nucleotide analog incorporated as part of a nucleotide sequence defining said labeled probe molecules.

C7

35. (Amended) The substrate of claim 31 whereby the incorporated nucleotide analog is 2-aminopurine replacing adenosine or guanine nucleotides.

38. (New) The substrate of claim 1 wherein the labeled probe fluoresces at a wavelength of about 300 nm to about 700 nm.

C8

39. (New) The method of claim 1 whereby the incorporated nucleotide analog is 2-aminopurine replacing at least one endemic adenosine or guanine nucleotide.

40. (New) The method of claim 20 whereby after incorporation of the nucleotide analog including 2-aminopurine, the labeled probe is affixed on a solid substrate.

I. REMARKS

Applicant's attorney wishes to thank the Examiner for her informative and helpful discussions regarding the instant application and, in particular, the references cited in the previous actions.

Claims 1, 6, 16, 17, 19, 20, 22-27, 29-31 and 35 have been amended to more particularly point out and distinctly claim certain aspects of Applicant's invention and in light of said discussions. The amended claims do not introduce new subject. Claims 2, 28, 33 and 36 have been cancelled without prejudice or disclaimer and new claims 38-40 are added.

Reconsideration and allowance of all of the claims in view of the above amendments and the following remarks are respectfully requested.

II. SYNOPSIS OF THE PRESENT INVENTION

Once again, Applicant thanks the Examiner for her time and effort discussing the instant case. It appears that throughout prosecution, much discussion has taken place regarding the various meanings and definitions of terms utilized both in the pending application and in the cited references.

In an attempt to clarify the germane issue that is crisply presented at this time and in this amendment, Applicant is using the terms "incorporated" nucleotide analogs in the claims now amended and submitted herein. This term has been essentially suggested by the examiner and applicant concurs that this recitation sets the invention clearly apart from the prior art. The term therein incorporated is not meant to imply attachment to nucleotide(s), such as nucleotide tags (i.e fluorescein labeling), but rather incorporation into the nucleic acid strand proper, that is, within or at one or both ends of a nucleic acid sequence.

The present invention provides an assay system using labeled probe molecules by incorporation of nucleotide analogs to identify and quantify unlabeled and unmodified target molecules in solution or sample. In one embodiment, labeled probe molecules are present on a solid support or substrate such as a microarray, and upon contacting the labeled probe with an unlabeled/unmodified target in solution, the identification of multiple different unlabeled/unmodified target molecules is made. In another embodiment, the incorporation of fluorescent nucleotide analogs into single stranded probes provides a first fluorescence which is reduced or quenched when said single stranded probes are paired or hybridized with unlabeled or unmodified target molecules, for example.

III. REJECTIONS UNDER 35 U.S.C. § 102

In the final office action, the examiner has rejected claims 1, 2 and 22-31 as being anticipated by U.S. Patent No. 6,100,030 to McCasky et al. (McCasky et al.). These rejections are respectfully traversed.

McCasky et al. does not disclose or suggest labeling probes by incorporation of nucleotide analogs and affixing the labeled probe on a substrate. McCasky et al. additionally does not teach a method for monitoring hybridization between an probe, having a nucleotide analog molecule that is incorporated and fluoresces, with complementary unlabeled target molecules, the monitoring or detection accomplished by the comparison of a first fluorescent level (the probe alone) and a second level of fluorescence (probe and hybridizing, unlabeled target), as recited in amended claims 1 and 22-24, for example. Missing elements cannot be supplied by the knowledge of one skilled in the art or the disclosure of another reference in order to give rise to an anticipation rejection. Structural Rubber Products Co. v. Park Rubber Co., 749 F.2d 707, 223 U.S.P.Q. 1264 (Fed. Cir. 1984).

Recitation of incorporated nucleotide analog which fluoresce, into probe molecules disposed onto a substrate as well as methods that utilize such a structure, is now recited in the pending claims. For example, McCasky et al. does not disclose labeled probe molecules having incorporated nucleotide analogs that fluoresce and whose florescence is utilized to measure or detect presence or hybridization of complementary molecules by quenching, or wherein the labeled probe fluoresces at a wavelength of about 300 nm to about 700 nm or wherein the incorporated nucleotide analog is 2-aminopurine replacing at least one endemic adenosine or guanine nucleotide (as recited in amended claim 1, new claims 38, 39 and pending claim 37, for example).

Accordingly, the absence of any teaching in McCasky et al. of probes having at least one nucleotide analog that fluoresces, the probes affixed on a substrate, demonstrates the fact that there is no anticipation. Furthermore, the methods of detection discussed and taught by McCasky et al. are related to elements that are not nucleotide analogs but rather **bound tags labels** (not incorporated) such as alkaline phosphatase molecules or horseradish peroxidase molecules, for example (col. 17 lines 49-50). The Applicant respectfully requests that the Examiner withdraw the anticipation rejections.

IV. REJECTIONS UNDER 35 U.S.C. § 103

The Examiner has rejected claims 4, 5, 10-13, 16-17, 19-20 and 32 as being unpatentable over McCasky et al. in view of McGall et al. These rejections are respectfully traversed.

In the present invention Claim 4 is drawn to a labeled probe comprised of native and nonnative nucleotides. Claim 5 is drawn to nucleotides being nucleotide analogs and recites several embodiments. Claims 10-12 are drawn to microarrays being divided into quadrants wherein each different quadrant has labeled probe molecules of different sequences. Claim 13 is drawn to a microarray being a bead. Amended claims 16-17 and 19-20 are drawn to methods comprising detecting a difference in fluorescence, provided by at least one incorporated nucleotide analog, before and after hybridization to a target molecule. Claim 32 is drawn to the method of Claim 16, wherein multiple labeled probes and the multiplying of the labeled probes are achieved by non-amplification steps.

In the final office action, the Examiner concedes that McCasky et al. ***does not teach*** nucleotide analogs (Claim 5), or arrays divided into quadrants having different labeled probes (Claims 10-12), or methods wherein the levels of label are expressed twice and compared (Claims 16-17, 19-20, and 22-25), or probes achieved by non-amplification steps (Claim 32) or the amount of probes on a substrate. Applicant notes once more that there is in particular no teaching regarding probes on a substrate having therein incorporated nucleotide analogs that fluoresce and whose florescence is utilized to measure or detect presence or hybridization of complementary molecules by quenching (claims 10-15 now depending, directly or indirectly from amended claim 1).

Furthermore, McCasky et al. teaches that "...hybridization is typically detected performed by monitoring a color shift resulting from the proximity of the two **bound** labels..." (col. 23, lines 49-50). The use of this term clearly shows that the labels taught and considered by McCasky et al. are limited to labels that are **bound** to DNA, that is, **tags** such as horseradish peroxidase (HRP) and/or alkaline phosphatase (AP), etc.. (col. 29, lines 42-55) and not to labeled probe molecules, said labeled probe molecules having incorporated nucleotide analogs that fluoresce and whose florescence is utilized to measure or detect presence or hybridization of complementary molecules, as presently claimed. The fluorescence of the probes of the present invention do not require a tagged marker or group bound to the DNA for visualization purposes, but it is the nucleotide analogs themselves which provide a user with a way to detect complementation/hybridization between affixed probe and the target.

Applicant has carefully considered the disclosure of McGall et al. and note that this reference teaches away from the methods and apparatus disclosed by the present invention. As opposed to labeling probes, as taught by the present application, the McGall et al. disclosure clearly teaches away and discloses the labeling of the target molecules and **not probes** (Field of the Invention). Further support for this assertion may be found in col. 10, lines 52-53, where "The nucleotide analogues are preferably incorporated into **target nucleic acids**..", which diametrically opposes the teachings of the present invention, as specified and detailed in the present claims.

Also, at col. 12 line 40, McGall et al. clearly teaches the labeling of a target with fluorescein and **does not** teach detection by labeling probe molecules, as stated by the Examiner on page 6 of the final office action. This further attests to the novelty and unobviousness of the presently claimed invention.

Additional disclosure which teaches away from the present invention may be found in col. 20 lines 16-20, which detail the fact that indeed, not only are targets utilized for assessing hybridization, but that the particular method of labeling utilizes 5'-fluorescein labeled oligo targets and not nucleotide analogs which fluoresce, as presently claimed in instant application. Furthermore, nowhere in McGall et al. is the incorporation of at least one nucleotide analogs that fluoresces, into probes, taught in order to utilize the florescence therefore provided to detect hybridization of target molecules. As stated previously, the targets in McGall et al. are labeled, and not even by the utilization of incorporated nucleotide analogs. Furthermore, the analogs in McGall et al. are disclosed for hydrolysis resistance or degradation by nuclease enzymes and further, in order to optimize mismatch discrimination and duplex stability (col. 2, lines 63-65 and col. 7 lines 35 and 36) and not for florescence which is utilized to measure or detect presence or hybridization of complementary molecules by quenching or a method for quantifying the amount of a target molecule that is itself unlabeled, as recited in the pending claims.

Clearly, in each of the above rejections under 35 U.S.C. §103, none of the references cited teach or suggest combining to produce the invention as claimed. Absent some teaching or suggestion to support the combination of the references, such combinations cannot establish obviousness. ACS Hospital Sys., Inc. v. Montefiore Hospital, 732 F.2d 1572, 1577 (Fed. Cir. 1984); In re Geiger, 815 F.2d 686, 788 (Fed. Cir. 1987); The desirability of combining two

references must be shown; it is not enough to merely state that such a combination is feasible. Winner Int'l Royalty Corp. v. Wang, 202 F.3d 1340 (Fed. Cir. 2000).

There is no suggestion or teaching in either the McCasky et al. or McGall et al. patents to combine the divergent teachings contained in each. There is no suggestion in either patent to combine a probe having incorporated therein a nucleotide analog that fluoresces to measure or detect presence or hybridization of complementary molecules by quenching or methods utilizing such a composition, in an array format. More particularly, there is no teaching or suggestion of utilizing the oligonucleotide arrays of McGall et al., which utilize nucleotide analogs in oligos having resistance to hydrolysis and optimized mismatch discrimination and duplex stability and which are hybridized to **labeled targets** (by utilizing 5'-fluorescein) which are they, themselves, utilized to detect hybridization, to the array of McCasky et al., wherein no probe is taught that has at least one nucleotide analog incorporated therein and that fluoresces and is used measure or detect presence or hybridization of complementary molecules by quenching, but rather to **bound labels** such as HRP or AP.

To the extent that the cited references be combined, the results would provide labeled targets (opposite to the teachings of the instant application) that are then hybridized to labeled oligos on an array, which are provided solely for their hydrolysis resistance or degradation by nuclease enzymes and further, in order to optimize mismatch discrimination and duplex stability (not nucleotide analogs which fluoresce and whose florescence is utilized to detect hybridization of target molecules, such as unlabeled targets, by quenching upon hybridization with the target). This, as described above, teaches away from the substrate and methods presently claimed in the instant application. Furthermore, the Examiner's stated reason for the obviousness of combining McCasky et al. and McGall et al., because McGall teaches that "oligonucleotide analogues are resistant to hydrolysis or degradation..." does not suggest or teach a method wherein oligonucleotide analogues that fluoresce are incorporated into a probe disposed upon a substrate and the use of florescence emanating from a said probe to detect hybridization of a target, including unlabeled targets, as taught by the present disclosure.

The Examiner has stated that it would have been obvious to one of ordinary skill to separate arrays of microarrays into different quadrants having different probes. The teachings of McGall et al. cited by the Examiner, namely col. 15 lines 55-59, are concerning the physical

locations of **unlabeled probes** on a chip, not probes having therein incorporated nucleotide analogs that fluoresce and whose quenching is utilized to detect hybridization. McGall et al. teaches that "hybridization is detected by **labeling a target** with e.g., fluorescein or other known visualization agents and incubating the target with an array of oligonucleotide probes (column 12, lines 39-42)." This is in **complete contrast** to the present invention, which claims methods having **labeled probes, NOT targets.**

It is clear that 35 U.S.C. §103 requires an analysis of the claimed invention as a whole, i.e. an analysis of the claimed combination of elements, including each and every limitation encompassed by the pending dependent claims. Even where the claimed invention is comprised of individual components well known at the time of invention, "[w]hat must be found obvious to defeat the patent **is the claimed combination**." The Gillette Co. v. S.C. Johnson & Son Inc., 16 USPQ2d 1923, 1927 (Fed. Cir. 1990). It is impermissible to simply to engage in a hindsight reconstruction of the claimed invention, using the applicant's structure as a template and selecting elements from references to fill in the gaps. The ***references themselves*** must provide some teaching whereby the applicant's combination would have been obvious. In re Gorman, 18 USPQ2d 1885, 1888 (Fed. Cir. 1991). No such suggestion is present here with any the references cited by the Examiner. Additional references, Gelfland et al., Scholin et al., Heagy and Mandecki fail to remedy this combination deficiency of the primary references and thus do not teach or suggest the claimed invention. As stated by the Examiner in the final office action, page 12, Scholin et al. teaches utilizing a florescent reagent that binds to the **target** after the target and probe are complexed. This does not even remotely relate to labeled probe molecules having therein incorporated nucleotide analogs that fluoresce and whose florescence is utilized to measure or detect presence or hybridization of complementary molecules by quenching (claim 1 from which claim 7 depends). In fact, the reagent does not and is not bound to the probe but to the target, again teaching away from the methods and apparatus presently claimed. Where references teach away from the invention, they cannot serve as a predicate for unobviousness, see McGinley v. Franklin Sports, Inc., 60 USPQ 2d 1001 at p. 1010, and cases cited therein.

As the cited references fail to suggest the combination of elements recited in the pending claims, whether viewed alone or in combination, Applicant respectfully submits that the pending claims are in condition for allowance, and the rejections under 35 U.S.C. §103 should be withdrawn.

In view of the above, it is submitted that this application is now in good order for allowance, and such early action is respectfully solicited. Should matters remain which the Examiner believes could be resolved in a telephone interview, the Examiner is requested to telephone the Applicant's undersigned attorney.

Respectfully submitted,

Dated: June 21, 2002

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ADDENDUM PAGES

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VERSION MARKED TO REFLECT CHANGES

ALL PENDING CLAIMS SHOWN

CHANGES IN THE CLAIMS

1. (Amended) A substrate having a surface area, the surface area comprising attached labeled probe molecules, said labeled probe molecules having therein incorporated nucleotide analogs that fluoresce and whose florescence being for measuring or detecting presence or hybridization of complementary molecules by quenching a first florescence provided by the labeled probe molecules.

4. The substrate of claim 1 wherein the labeled probe is comprised of native and nonnative nucleotides.

5. The labeled probe molecules of Claim 1 wherein the nucleotides are nucleotide analogs including 2-amino purine for adenosine or guanine; ribonucleoside or 2,6-diamino ribonucleoside, formycin A, formycin B, oxyformycin B, toyocamycin, sangivamycin, pseudoouridine, showdomycin, minimycin, pyrazomycin, 5-amino-formycin A, 5-amino-formycin B or 5-oxo-formycin A [at least] for adenosine; 4-amino-pyrazolo [3,4d] pyrimidine, 4,6-diamino-pyrazolo [3,4d] pyrimidine, 4-amino-6-oxo-pyrazolo [3,4d] pyrimidine, 4-oxo-pyrazolo [3,4d] pyrimidine, 4-oxo-6-amino-pyrazolo [3,4d] pyrimidine, 4,6-dioxo-pyrazolo [3,4d] pyrimidine, pyrazolo [3,4d] pyrimidine, 6-amino-pyrazolo [3,4d] pyrimidine or 6-oxo-pyrazolo [3, 4d] pyrimidine for cytosine or thymidine.

6. (Amended) The labeled probe molecules of claim [2] 1, wherein the nucleotide analog is 2-amino purine.

7. The substrate of Claim 1 wherein the labeled probe molecules are comprised of amino acids.

10. The substrate of Claim 1 wherein the substrate is a microarray further having the surface area divided into quadrants wherein each different quadrant has different labeled probe molecules.

11. The microarray substrate of claim 10 having from about 100 to about 10,000 different labeled probe molecules located upon about 100 to about 10,000 different quadrants.

12. The microarray of claim 10 having about 100 to about 1,000 labeled probe molecules per quadrant.

13. The substrate of Claim 1 wherein the substrate is a bead, said bead sizes range from about 10 microns to about 20 microns.

14. The bead substrate of claim 13 wherein the bead is formed of a ferromagnetic metal core and a polymeric coating.

15. The bead substrate of claim 13 having from about 100 to about 1,000 labeled probe molecules attached to the surface area of the bead.

16. (Amended) A method for assessing the presence of a target molecule in a cell or tissue sample comprising the steps of:

a. [procuring] providing a microarray having a surface area comprising attached labeled probe molecules in quadrants, said labeled probe molecules including at least one nucleotide analog capable of fluorescence;

b. detecting [the level of label] fluorescence from said at least one nucleotide analog capable of fluorescence expressed within [each] quadrants a first time;

c. applying a sample comprising unlabeled target sequences to the microarray;

d. providing a sufficient condition[s] and time for target molecules to selectively pair with [the] complementary labeled probe molecules; [and]

e. detecting [the level of label] fluorescence from said at least one nucleotide analog capable of fluorescence expressed within [each] quadrants a second time;

f. comparing the [levels of label] fluorescence expressed between the first time and the second time for each quadrant[.];

g. repeating steps c - f until [the] levels of fluorescence [label] approach[es] zero and/or about background levels; and

h. the difference between [levels] fluorescence [of label] in that of step f and that of step c [identifies] identifying a target/probe pair.

17. (Amended) A method for quantifying the amount of a target molecule in solution comprising the steps of:

a. [procuring] providing a first substrate having a surface area comprising a known number of labeled probe molecules, said labeled probe molecules include at least one nucleotide analog capable of fluorescence;

b. detecting a first level of nucleotide analog fluorescence [label] expressed by the labeled probe molecules on the first substrate;

c. contacting the first substrate with a volume of sample containing unlabeled target nucleotide sequences;

d. providing a sufficient condition[s] and time for unlabeled target molecules to selectively pair with the labeled probe molecules;

e. removing the first substrate and detecting the level of nucleotide analog fluorescence [label] expressed by [the substrate] said known number of labeled probe molecules after exposure to the sample containing unlabeled target molecules;

f. where the level of nucleotide analog fluorescence [label] expression of the first substrate is substantially reduced to levels substantially similar to background levels, repeating steps a. through e. with subsequent substrates, having surface areas comprising known numbers of labeled probe molecules; and [.]

g. calculating the amount of target molecule in the volume of sample by adding the known number of labeled probe molecules present on the first substrate and subsequent substrates contacted with the sample, wherein the levels of nucleotide analog fluorescence [label] expression of the substrates [were] are reduced relative to the levels prior to contacting the sample.

18. The method of claim 10, wherein the level of label expression is evaluated using a flow cytometer.

19. (Amended) A substrate having a surface area divided into quadrants comprising:[;]

different nucleotide probe molecule sequences bound to the surface area, wherein different nucleotide probe molecule sequences are bound to distinct quadrants;

[wherein] the nucleotide probe molecules [are characterized as] being a single stranded form or double stranded [in] form, said nucleotide probe molecules having incorporated nucleotide analogs that fluoresce, wherein the level of label expressed from the single stranded probe molecules is greater than the level of label expressed from the double stranded probe molecules; and

wherein the nucleotide probe molecules [are further characterized by] have an ability to hybridize to target nucleotide sequences.

20. (Amended) A method for monitoring the hybridization of target and probe by complementation, [said method] comprising [of]:

a. incorporating fluorescent nucleotide analogs [molecules] into probes;

b. detecting a first level of [label in] fluorescence emanating from probes of step a;

d. hybridizing a target with said [labeled] probes thereby forming a probe-target complex;

e. detecting a second level of [label] fluorescence emanating from said probe-target complex after hybridization of probe and target;

f. comparing the first and second levels of [label] fluorescence between that of step b and that of step e, and wherein said difference between second and first levels is less than said first level of step b;

g. washing of unhybridized target; and

h. repeating steps d - g until the difference between the first and second levels of fluorescence [label] approaches approximately zero and/or about background levels.

22. (Twice Amended) A method for monitoring the hybridization of a probe and a target comprising, a fluorescently labeled probe, said fluorescence being provided by a nucleotide analog capable of fluorescence and is incorporated, thereby providing a detectable first level of fluorescence and providing a detectable second level of fluorescence when the labeled probe is hybridized to a complementary unlabeled target, wherein the second level is lower than the first level.

23. (Twice Amended) A method for monitoring the hybridization of a probe and a target comprising supplying a fluorescently labeled probe, said fluorescently labeled probe being fluorescent due to the incorporation of at least one nucleotide analog thereby providing a detectable first level of fluorescence, and providing a detectable second level of fluorescence when the labeled probe is hybridized to a complementary unlabeled target, wherein the second level is significantly lower than the first level.

24. (Twice Amended) A method for monitoring the hybridization of a probe and a target comprising supplying a fluorescently labeled probe, said fluorescently labeled probe being fluorescent due to the incorporation of at least one nucleotide analog capable of fluorescence, thereby providing a detectable first level of fluorescence, and providing a detectable second level of fluorescence when the labeled probe is hybridized to a complementary unlabeled target, wherein the second level is approximately zero.

25. (Twice Amended) A method for monitoring the hybridization of a probe and a target comprising supplying a fluorescently labeled probe, said fluorescently labeled probe being fluorescent due to the incorporation of at least one nucleotide analog capable of fluorescence, thereby providing a detectable first level of fluorescence, and a detectable second level of fluorescence when the labeled probe is hybridized to a complementary unlabeled target, wherein the second level is approximately zero and the first level is greater than zero.

26. (Amended) A substrate having a plurality of probes, wherein said probes are fluorescently labeled, said fluorescently labeled probes include incorporated nucleotide analogs that fluoresce and whose florescence is utilized to measure or detect presence or hybridization of

complementary, unlabeled molecules, the labeled probe providing a detectable first level of fluorescence.

27. (Twice Amended) A substrate having a plurality of probes, wherein said probes are fluorescently labeled by incorporation of at least one nucleotide analog, the labeled probe providing a detectable first level of fluorescence, and when hybridized to a complementary target providing a second level of fluorescence, wherein the second level is lower than the first level and said levels of fluorescence being derived from excitation of said at least one nucleotide analog.

29. (Twice Amended) A substrate having a plurality of probes, wherein said probes are fluorescently labeled by incorporation of at least one nucleotide analog, the labeled probe providing a detectable first level of fluorescence, and when hybridized to a complementary target having no nucleotide analogs incorporated therein, providing a second level of fluorescence, wherein the second level approaches zero.

30. (Twice Amended) A substrate having a plurality of probes, wherein said probes are fluorescently labeled by incorporation of at least one nucleotide analog, the labeled probe providing a detectable first level of fluorescence, and when hybridized to a complementary target having no nucleotide analogs incorporated therein providing a second level of fluorescence, wherein the second level is greater than zero, said levels of fluorescence being derived from excitation of said at least one nucleotide analog.

31. (Amended) A substrate having a surface area, the surface area comprising attached labeled probe molecules, said probe further comprising a fluorescent label, said fluorescent label including at least one nucleotide analog incorporated as part of a nucleotide sequence defining said labeled probe molecules.

32. The method of claim 16, 17, 18, 20, 22, 23, 24 or 25, wherein the multiple labeled probes and the multiplying of the labeled probes are achieved by a non-amplification step.

34. The method of claim 31 whereby the labeled probe molecules are nucleotide analogs including 2-amino purine for adenosine or guanine; ribonucleoside or 2,6-diamino ribonucleoside, formycin A, formycin B, oxyformycin B, toyocamycin, sangivamycin, pseudoouridine, showdomycin, minimycin, pyrazomycin, 5-amino-formycin A, 5-amino-

formycin B or 5-oxo-formycin A for adenosine; 4-amino-pyrazolo [3,4d] pyrimidine, 4,6-diamino-pyrazolo [3,4d] pyrimidine, 4-amino-6-oxo-pyrazolo [3,4d] pyrimidine, 4-oxo-pyrazolo [3,4d] pyrimidine, 4-oxo-6-amino-pyrazolo [3,4d] pyrimidine, 4,6-dioxo-pyrazolo [3,4d] pyrimidine, pyrazolo [3,4d] pyrimidine, 6-amino-pyrazolo [3,4d] pyrimidine or 6-oxo-pyrazolo [3, 4d] pyrimidine for cytosine or thymidine.

35. (Amended) The [method] substrate of claim 31 whereby the incorporated nucleotide analog is 2-aminopurine replacing adenosine or guanine nucleotides.

37. The method for quantifying the amount of a target molecule in solution comprising the steps of:

a. incorporating a nucleotide analog including 2-aminopurine into a probe;

b. affixing the labeled or modified probe on a substrate;

c. detecting a first level of label expressed by the labeled or modified probe molecules on the substrate;

d. contacting substrate with a volume of sample containing unlabeled or unmodified target molecules in solution;

e. providing sufficient conditions and time for unlabeled or unmodified target molecules in solution to selectively pair and hybridize with the labeled probe molecules affixed on the substrate;

f. removing the substrate and detecting the second level of label expressed by the labeled probed affixed on the substrate after exposure to the unlabeled or unmodified target molecules in solution;

g. comparing the first and second levels of label expressed by the labeled or modified probe;

h. identifying probe and target hybridized molecules by repeating steps c-f until the amounts of label expression between the first and second levels of label approaches zero and/or about background levels.

38. (New) The substrate of claim 1 wherein the labeled probe fluoresces at a wavelength of about 300 nm to about 700 nm.

39. (New) The method of claim 1 whereby the incorporated nucleotide analog is 2-aminopurine replacing at least one endemic adenosine or guanine nucleotide.

40. (New) The method of claim 20 whereby after incorporation of the nucleotide analog including 2-aminopurine, the labeled probe is affixed on a solid substrate.

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